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A).

1 MALWMRLPL LALLALWGPD PAAAFVNGHL CGSHLVEALY LVCGERGFFY  
51 TPKTRREAED LQVGQVELGG GPGAGSLQPL ALEGLGKRG IVEQCCTSIC  
101 SLYQLENYCN

B).

A-chain

B-chain

1 GIVEQCCTSI CSLYQLENYC NFVNQHLGSH HLVEALYLVC GERGFFYTPK T

C).

1TRZ:A

1 GIVEQCCTSI CSLYQLENYC N  
HHHHTTS HHHHGGG

1TRZ:B

1 FVNQHLGSH LVEALYLVC GERGFFYTPKT  
S HHH HHHHHHHHHG GG

1TRZ:C

1 GIVEQCCTSI CSLYQLENYC N  
HHHHSSS HHHHGGG

1TRZ:D

1 FVNQHLGSH LVEALYLVC GERGFFYTPKT  
HHHHHHH HHHHHHHHHG GG S

Figure 1

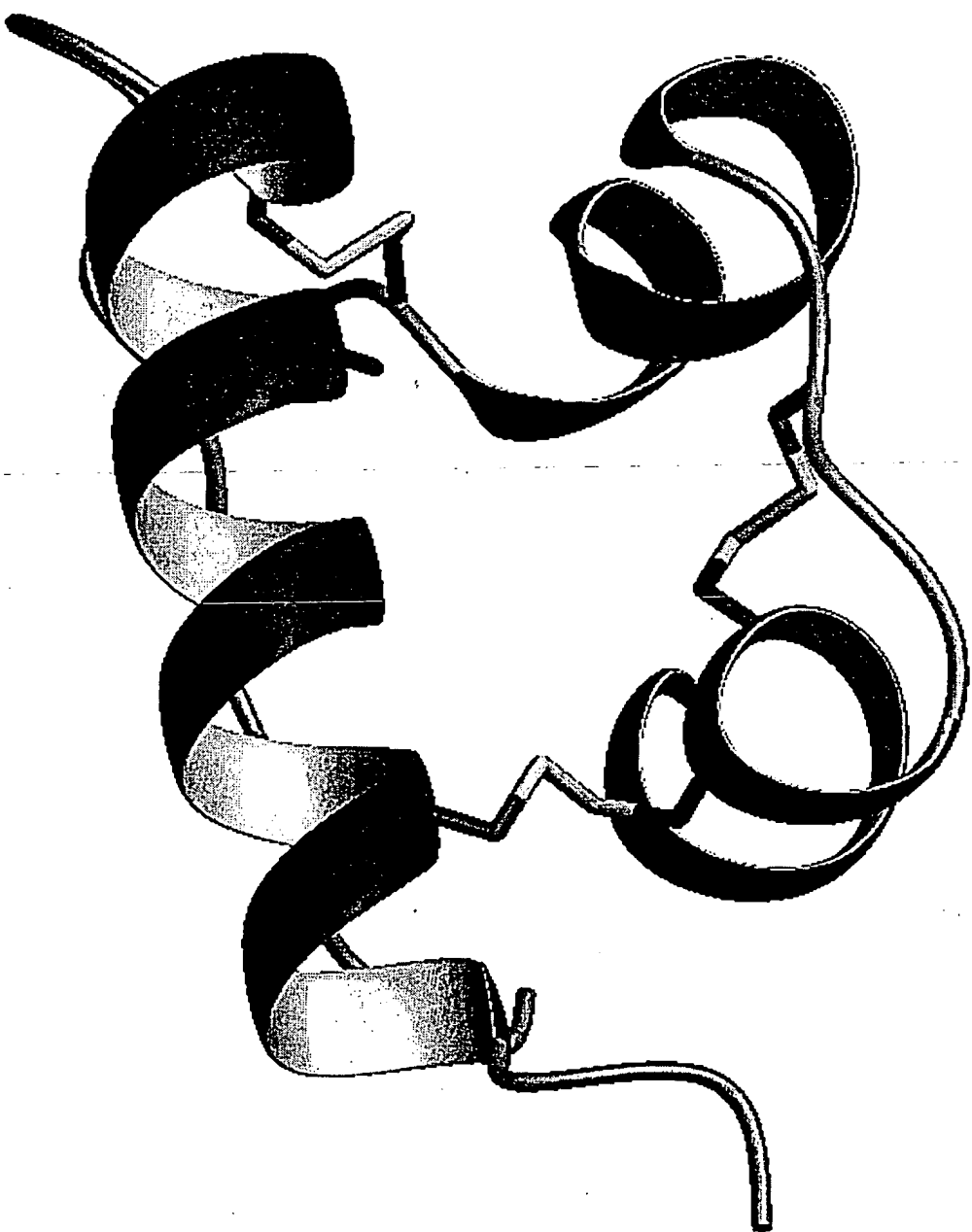


FIGURE 2A



**FIGURE 2B**

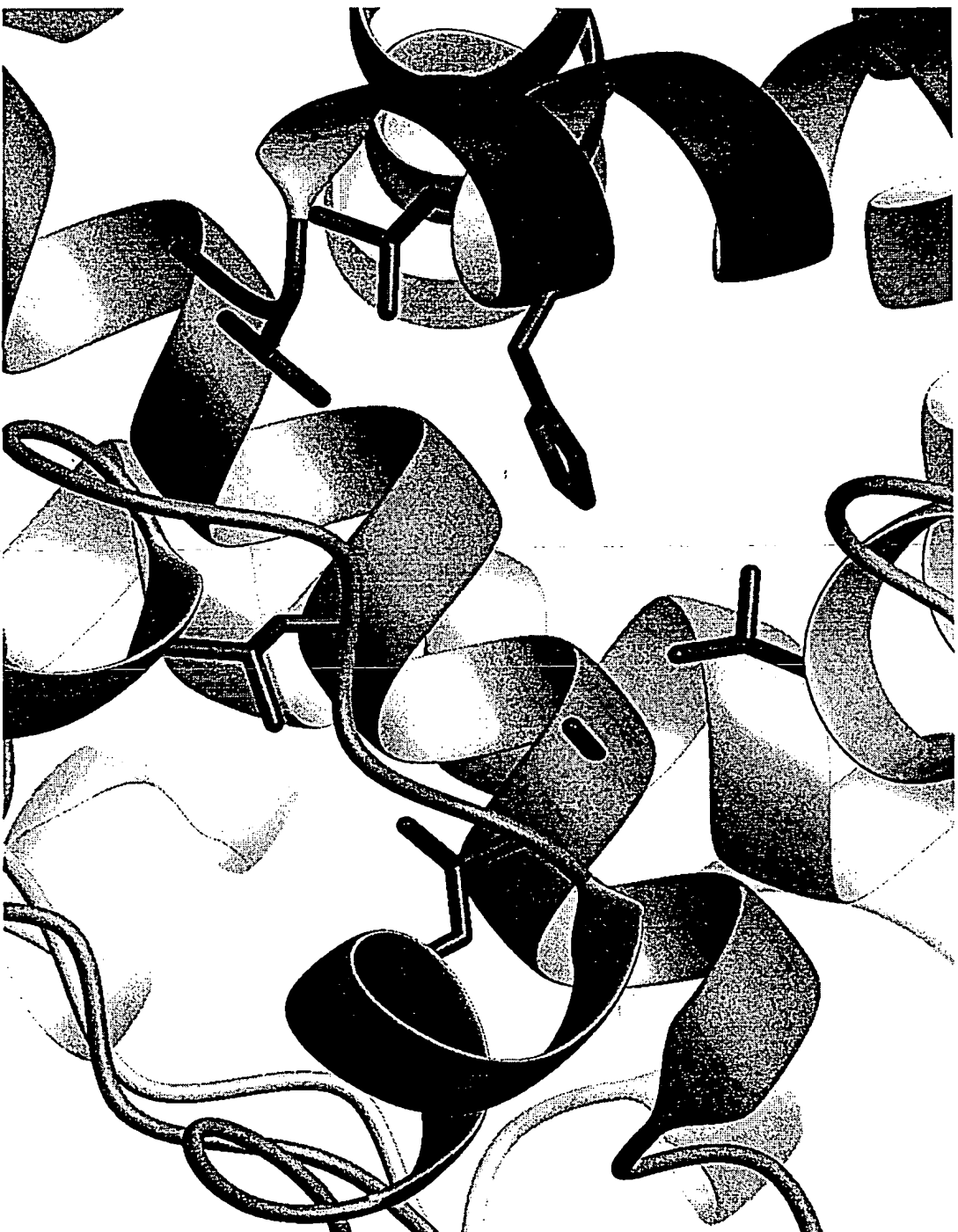


FIGURE 2C

A). A-chain	B-chain
1 NLVEQASTSQ ASLYQINFD NDVNEFHLYGS HIREWLYLVA GERGFNFDPK T

B). A-chain	B-chain
1 GIVEQCSTSI CSLYQLENYC NFENYHLYGS HLVEALYLVC GERGFFYTPK T

C). A-chain	B-chain
1 GIVEQCSTSI CSLYQLENYC NFNQHLDGS HLVEALYLVC GERGFFYTPK T

D). A-chain	B-chain
1 GIVEQCSTSI CSLYQLENYC NFTNYHLYGS HLVEALYLVC GERGFFYTPK T

E). A-chain	B-chain
1 GIVEQCSTSI CSLYQLENYC NFNVNYHLYGS HLVEALYLVC GERGFFYTPK T

F). A-chain	B-chain
1 GIVEQCSTSI CSLYQLENYC NFNQHLEGS HLVEALYLVC GERGFFYTPK T

G). A-chain	B-chain
1 GIVEQCETSI CSLYQLENYC N----HLEGS HLVEALYLVC GERGFFYTPK T

### Figure 3

A).                    **A-chain**                    **B-chain**  
 -----|-----  
 1 GIVEQCCTSI CSLYQLENYC NFNQFLCGS HLVEFLYLVLC GERGFFYTPK T

B).                    **A-chain**                    **B-chain**  
 -----|-----  
 1 GIVEQCCTSI CSLYQLENYC NFNQFLCGS HLVEALYLVLC GERGFFYTPK T

C).                    **A-chain**                    **B-chain**  
 -----|-----  
 1 GIVEQCCTSI CSLYQLENYC NFNQHLCGS HLVEFLYLVLC GERGFFYTPK T

D).                    **A-chain**                    **B-chain**  
 -----|-----  
 1 GIVEQCCTSI CSLYQLENYC NFNQHLCGS HLVEWLYLVLC GERGFFYTPK T

E).                    **A-chain**                    **B-chain**  
 -----|-----  
 1 GIVEQCCTSI CSLYQLENYC NFNQFLCGS HLVEWLYLVLC GERGFFYTPK T

F).                    **A-chain**                    **B-chain**  
 -----|-----  
 1 GIVEQCCTSI CSLYQLENYC NFNQHLCGS HLVEXLYLVLC GERGFFYTPK T

G).                    **A-chain**                    **B-chain**  
 -----|-----  
 1 GIVEQCCTSI CSLYQLENYC NFNQHLCGS HLVEILYLVLC GERGFFYTPK T

**Figure 4**

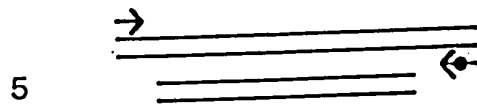
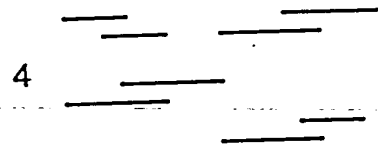
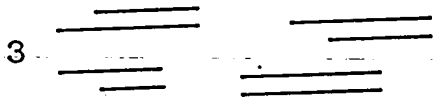
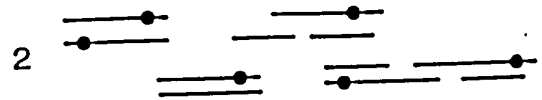
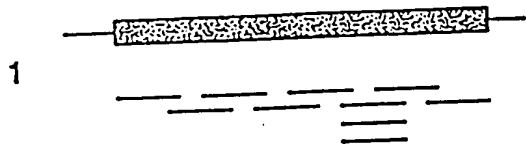
A).                      **A-chain**                      **B-chain**  
 -----|-----  
 1 NIVEQCCTSQ CSLYQYENYC NDVNQHLCGS HLVEALYLVC GERGFNYDPK T

B).                      **A-chain**                      **B-chain**  
 -----|-----  
 1 NIVEQCCTSQ CSLYQYYNFC NDKNFHLCGS HIREWLYLVC GERGFNFDPK T

C).                      **A-chain**                      **B-chain**  
 -----|-----  
 1 NIVEQCCTSQ CSLYQYYNFC NDKNFHLCLS HIREWLYLVC GERGFNFDNK T

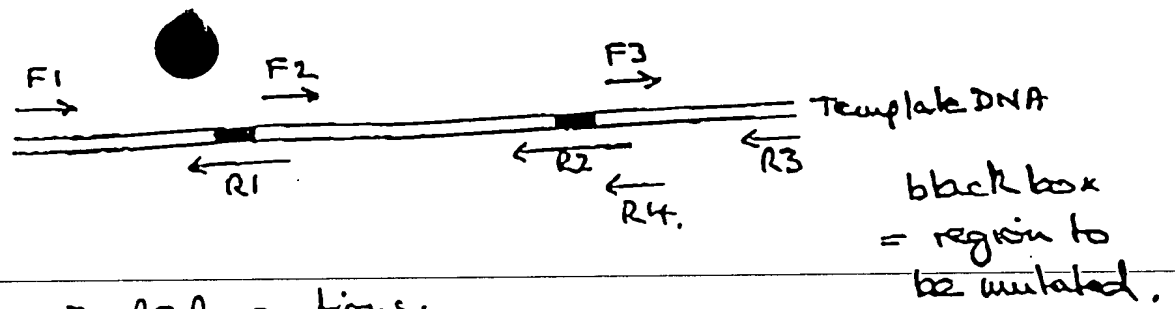
**Figure 5**





**FIGURE 6**

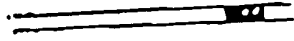




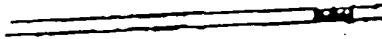
Step 1: Set up 3 PCR reactions.

Products:

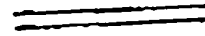
Tube 1:



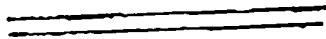
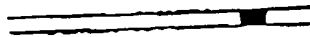
Tube 2:



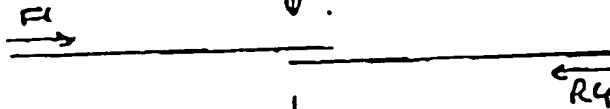
Tube 3:



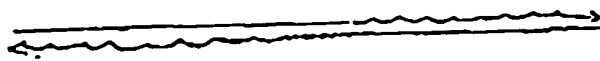
Step 2: Set up PCR reaction with products of tube 1 + products tube 2 + F1 + R4



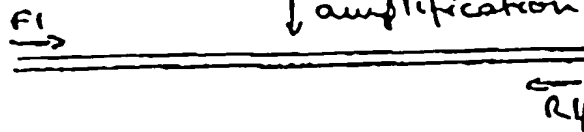
↓ Heat + anneal phase of PCR,



↓ synthesis phase of PCR.



↓ amplification phase, using F1 + R4.



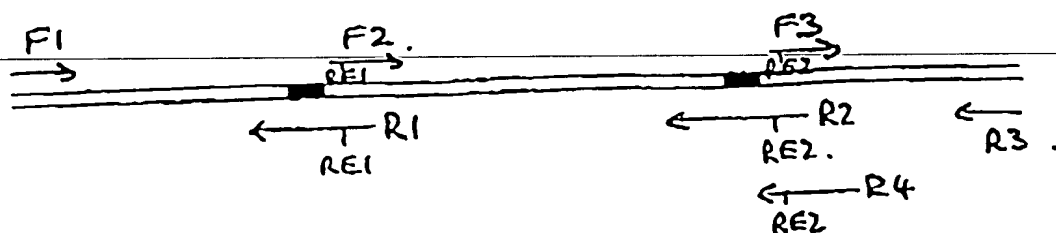
during subsequent cycles.

FIGURE 8A

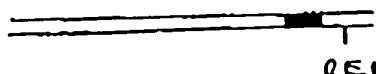
Step 3.

Repeat step 2 using product from step 2 +  
product from Step 1, tube 3 + primers F1 + R3

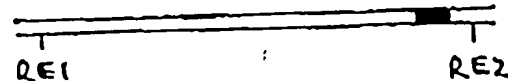
**FIGURE 8B**



Step 1 Set up 3 PCR reactions:

Tube 1: 

The diagram for Tube 1 shows a horizontal line representing a DNA segment. A vertical tick mark labeled RE1 is at the right end of the segment.

Tube 2: 

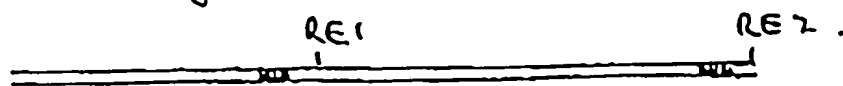
The diagram for Tube 2 shows a horizontal line representing a DNA segment. Vertical tick marks labeled RE1 and RE2 are at the left and right ends of the segment, respectively.

Tube 3: 

The diagram for Tube 3 shows a horizontal line representing a DNA segment. A vertical tick mark labeled RE2 is at the right end of the segment.

Step 2: digest products from Step 1 with suitable restriction endonucleases

Step 3: ligate digested product from Step 2, Tube 2 with digested product from Step 2, Tube 1.



**FIGURE 9A**

Step 4

Amplify ligated products of Step 3 with F1 + R4.  
via PCR

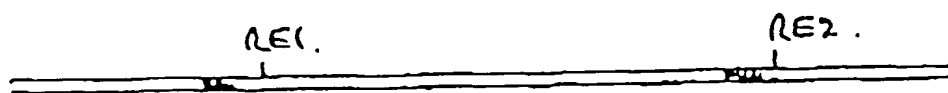


Step 5. Digest amplified product of step 4 with restriction: endonuclease #2.



Step 6.

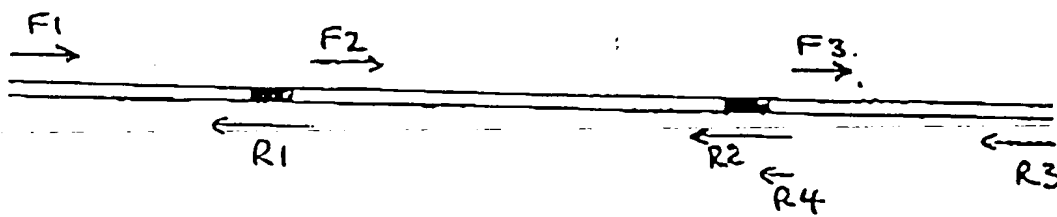
Ligate product from Step 5 with product from Step 2, tube 1.



Step 7

Amplify product from step 6 with F1 + R3

FIGURE 9B



**FIGURE 10**